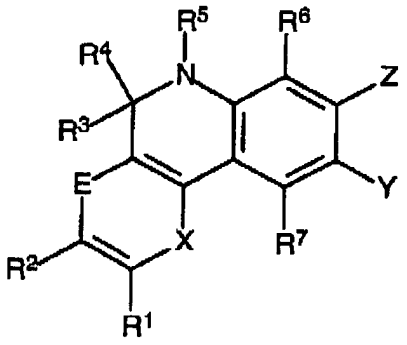
1. (Amended) A compound [of the] comprising a formula



wherein

R^1 and R^2 are independently selected from the group consisting of [H] hydrogen, cyano, halogen, carboxylic acid, [or] sulfonic acid[; or] . [a] C_1-C_6 alkyl, [or] C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy [that] is optionally substituted by carboxylic acid, sulfonic acid, or halogen[; or an aryl or heteroaryl ring] and said aryl or heteroaryl [that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl; [or -L--R_X; or -L-S_C;]

or R^1 in combination with R^2 forms a fused aromatic or heteroaromatic ring that is optionally sulfonated one or more times, or [which] said ring is substituted by -L-R_X or -L-S_C;

or R^2 in combination with R^3 forms a 5- or 6-membered alicyclic ring;

R^3 and R^4 are independently selected from the group consisting of [H] hydrogen, C_1-C_6 alkyl, aromatic or heteroaromatic ring, -L-R_X and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, hydroxy, or halogen[; or an aromatic or heteroaromatic ring] and said aromatic or heteroaromatic ring [that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 alkoxy, C_1-C_6

perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_X; or -L-S_C;]

or R^3 in combination with R^4 forms a 5- or 6-membered alicyclic ring;

R^5 is independently selected from the group consisting of [H] hydrogen, methyl, carboxymethyl, [a] C_2-C_6 alkyl, aryl, heteroaryl, -L-R_X and -L-S_C wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen[; or R^5 is an aryl or heteroaryl ring that] and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_X; or -L-S_C;]

R^6 is independently selected from the group consisting of [H] hydrogen, cyano, halogen, carboxylic acid, [or] sulfonic acid[; or a] , C_1-C_6 alkyl, [or] C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy [that] is optionally substituted by carboxylic acid, sulfonic acid, or halogen[; or an] and said aryl or heteroaryl [ring that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_X; or -L-S_C;]

or R^4 in combination with R^5, or R^5 in combination with R^6, forms a 5- or 6-membered alicyclic ring;

R^7 is independently selected from the group consisting of hydrogen, C_1-C_6 alkyl [having 1-6 carbons, or], C_1-C_6 alkoxy [having 1-6 carbons; or], -L-R_X[; or] and -L-S_C;

one of X and E is O, S, NR^8, or $CR^{1'}=CR^{2'}$, and the other is absent;

wherein R^8 is independently selected from the group consisting of [H] hydrogen, methyl, carboxymethyl, [or a] C_2-C_6 alkyl, -L-R_X and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; [or -L-R_X; or -L-S_C:] and

$R^{1'}$ and $R^{2'}$ are independently selected from the group consisting of [H] hydrogen, cyano, halogen, carboxylic acid, [or] sulfonic acid[; or] , [a] C_1-C_8 alkyl, [or] C_1-C_8 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy [that] is optionally substituted by carboxylic acid, sulfonic acid, or halogen[; or an aryl or heteroaryl ring] and said aryl or heteroaryl [that] is optionally substituted one or more times by C_1-C_8 alkyl, C_1-C_8 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_X or -L-S_C;]

Y is independently selected from the group consisting of H, OH, NH_2, NO, -(CO)-R^9, -(CO)-O-R^{10}, wherein said R^9 and R^{10} are independently H, C_1-C_8 alkyl, or a substituted or unsubstituted aryl or heteroaryl ring system having 1-2 rings;

Z is independently selected from the group consisting of H, OH, NHR^{17}, SH, or $C(CR^{11}R^{12})_2OH$; wherein said R^{17} is a C_1-C_8 alkyl that is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen[;] and said R^{11} and R^{12} are independently C_1-C_8 alkyl that are optionally substituted by carboxylic acid, sulfonic acid, or halogen, or R^{11} and R^{12} taken in combination form a 5- or 6-membered alicyclic ring;

wherein L is a covalent linkage;

R_x is a reactive group; and

S_C is a conjugated substance.

2. (Amended) [A] The compound[, as claimed in] according to Claim 1, wherein one of X and E is O, S, or $CR^{1'}=CR^{2'}$, and the other is absent.

3. (Amended) [A] The compound[, as claimed in] according to Claim [1] 2, wherein said compound [having the] has the formula

R^3 and R^4 are each methyl;

R^6 and R^7 are each hydrogen or methyl; and

Z is OH.

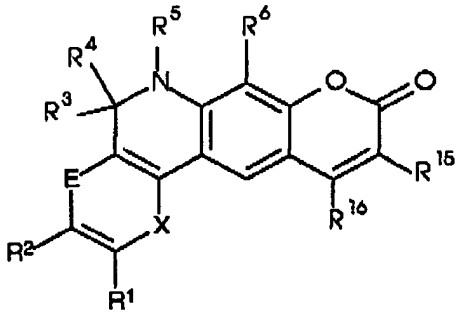
7. (Amended) [A] The compound[, as claimed in] according to Claim 1, wherein Y is H or -(CO)-H or NO.

8. (Amended) [A] The compound[, as claimed in] according to Claim 1, wherein [each] said L is independently a single covalent bond[,] or [L is] a covalent linkage having 1-2[4]10 nonhydrogen atoms selected from the group consisting of C, N, O, P, and S, [and is composed of any combination of single, double, triple or aromatic carbon–carbon bonds, carbon–nitrogen bonds, nitrogen–nitrogen bonds, carbon–oxygen bonds, carbon–sulfur bonds, phosphorus-oxygen bonds, and phosphorus-nitrogen bonds.]

9. (Amended) [A] The compound[, as claimed in] according to Claim 1, wherein said R_x is independently selected from the group consisting of an acrylamide, an activated ester of a carboxylic acid, an acyl azide, an acyl nitrile, an aldehyde, an alkyl halide, an amine, an anhydride, an aniline, an aryl halide, an azide, an aziridine, a boronate, a carboxylic acid, a diazoalkane, a haloacetamide, a halotriazine, a hydrazine, an imido ester, an isocyanate, an isothiocyanate, a maleimide, a phosphoramidite, a reactive platinum complex, a sulfonyl halide, [or] and a thiol group.

10. (Amended) [A] The compound[, as claimed in] according to Claim 1, wherein said S_c is independently selected from the group consisting of an amino acid, a peptide, a protein, a tyramine, a [monosaccharide, a polysaccharide] carbohydrate, an ion-complexing moiety, a nucleoside, a nucleotide, an oligonucleotide, a nucleic acid, a hapten, a psoralen, a drug, a hormone, a lipid, a lipid assembly, a polymer, a polymeric microparticle, a biological cell, [or] and a virus.

11. (Amended) A compound [of the] comprising a formula



wherein R^1, R^2, and R^6 are independently selected from the group consisting of [H] hydrogen, cyano, halogen, carboxylic acid, [or] sulfonic acid[; or a] C_1-C_6 alkyl, [or] C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_x and -L-S_C, wherein said alkyl or alkoxy [that] is optionally substituted by carboxylic acid, sulfonic acid, or halogen[; or an aromatic or heteroaromatic ring that] and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_x; or -L-S_C;]

or R^1 in combination with R^2 forms a fused aromatic or heteroaromatic ring that is optionally sulfonated one or more times, or said ring is substituted by -L-R_X or -L-S_C;

R^3 and R^4 are independently selected from the group consisting of [H] hydrogen, C_1-C_6 alkyl, an aromatic or heteroaromatic ring, L-R_x and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, hydroxy, or halogen[; or] and said [an] aromatic or heteroaromatic ring [that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 alkoxy, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_x; or -L-S_C;]

or R^2 in combination with R^3, or R^3 in combination with R^4, forms a 5- or 6-membered alicyclic ring;

R^5 is independently selected from the group consisting of [H] hydrogen, methyl, carboxymethyl, [a] C_2-C_6 alkyl, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen[; or R^5 is an] and said aryl or heteroaryl [ring that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_X; or -L-S_C;]

or R^4 in combination with R^5, or R^5 in combination with R^6, forms a 5- or 6-membered alicyclic ring;

one of X and E is O, S, NR^8, or $CR^{1'}=CR^{2'}$[;] and the other is absent;

wherein R^8 is independently selected from the group consisting of [H] hydrogen, methyl, carboxymethyl, [or a] C_2-C_6 alkyl, -L-R_X and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; [or -L-R_X; or -L-S_C;] and

$R^{1'}$ and $R^{2'}$ are independently selected from the group consisting of [H] hydrogen, cyano, halogen, carboxylic acid, [or] sulfonic acid[; or] , [a] C_1-C_6 alkyl, [or] C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy [that] is optionally substituted by carboxylic acid, sulfonic acid, or halogen[; or an aryl or heteroaryl ring] and said aryl or heteroaryl [that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_X; or -L-S_C;]

R^{15} and R^{16} are independently selected from the group consisting of hydrogen, cyano, nitro, halogen, carboxylic acid, [or] sulfonic acid[; or a], C_1-C_6 alkyl, an aromatic or heteroaromatic ring system having 1-2 fused rings, -L-R_X and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, or halogen[; or an] and said aromatic or heteroaromatic ring system [having 1-2 fused rings that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_X; or -L-S_C;]

wherein L is a covalent linkage;

R_x is a reactive group; and

S_C is a conjugated substance.

12. (Amended) [A] The compound[, as claimed in] according to Claim 11, wherein said one of X and E is O or S.

13. (Amended) [A] The compound[, as claimed in] according to Claim 12, wherein

R^6 and R^7 are [H] hydrogen;

R^3 and R^4 are each methyl;

R^1 is [H] hydrogen or sulfonic acid;

one of R^{15} and R^{16} is -L-R_x or -L-S_C, and the other is hydrogen, C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl; or cyano;

wherein L is a single covalent bond, or L is a covalent linkage having 1-2[4]0 nonhydrogen atoms selected from the group consisting of C, N, O, P, and S [and is composed of any combination of single, double, triple or aromatic carbon–carbon bonds, carbon–nitrogen bonds, nitrogen–nitrogen bonds, carbon–oxygen bonds, carbon–sulfur bonds, phosphorus-oxygen bonds, and phosphorus-nitrogen bonds], and wherein R_X[, when present,] is independently selected from the group consisting of an acrylamide, an activated ester of a carboxylic acid, an acyl azide, an acyl nitrile, an aldehyde, an alkyl halide, an amine, an anhydride, an aniline, an aryl halide, an azide, an aziridine, a boronate, a carboxylic acid, a diazoalkane, a haloacetamide, a halotriazine, a hydrazine, an imido ester, an isocyanate, an isothiocyanate, a maleimide,

wherein L is a covalent linkage;

R_x is a reactive group; and

S_c is a conjugated substance.

12. (Amended) [A] The compound[, as claimed in] according to Claim 11, wherein said one of X and E is O or S.

13. (Amended) [A] The compound[, as claimed in] according to Claim 12, wherein

R^6 and R^7 are [H] hydrogen;

R^3 and R^4 are each methyl;

R^1 is [H] hydrogen or sulfonic acid;

one of R^{15} and R^{16} is -L-R_x or -L-S_c, and the other is hydrogen, C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl; or cyano;

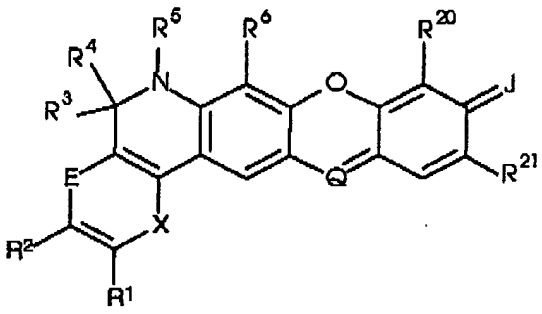
wherein L is a single covalent bond, or L is a covalent linkage having 1-2[4]0 nonhydrogen atoms selected from the group consisting of C, N, O, P, and S [and is composed of any combination of single, double, triple or aromatic carbon–carbon bonds, carbon–nitrogen bonds, nitrogen–nitrogen bonds, carbon–oxygen bonds, carbon–sulfur bonds, phosphorus-oxygen bonds, and phosphorus-nitrogen bonds], and wherein R_x[, when present,] is independently selected from the group consisting of an acrylamide, an activated ester of a carboxylic acid, an acyl azide, an acyl nitrile, an aldehyde, an alkyl halide, an amine, an anhydride, an aniline, an aryl halide, an azide, an aziridine, a boronate, a carboxylic acid, a diazoalkane, a haloacetamide, a halotriazine, a hydrazine, an imido ester, an isocyanate, an isothiocyanate, a maleimide,

a phosphoramidite, a reactive platinum complex, a sulfonyl halide, [or] and a thiol group; and

wherein S_C [, when present,] is independently selected from the group consisting of an amino acid, a peptide, a protein, a tyramine, a [monosaccharide, a polysaccharide] carbohydrate, an ion-complexing moiety, a nucleoside, a nucleotide, an oligonucleotide, a nucleic acid, a hapten, a psoralen, a drug, a hormone, a lipid, a lipid assembly, a polymer, a polymeric microparticle, a biological cell, [or] and a virus.

14. (Amended) [A] The compound[, as claimed in] according to Claim 11, wherein one of said R^{15} [and] or R^{16} is an aromatic or heteroaromatic ring system having 1-2 fused rings that is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl.

15. A compound [of the] comprising a formula:



[wherein]

wherein R^1, R^2, and R^6 are independently selected from the group consisting of [H] hydrogen, cyano, [nitro,] halogen, carboxylic acid, [or] sulfonic acid[; or a] C_1-C_6 alkyl, [or] C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_x and -L-S_C, wherein said alkyl or alkoxy [that] is optionally substituted by carboxylic acid, sulfonic acid, or halogen[; or an aromatic or heteroaromatic ring that] and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_x; or -L-S_C;]

or R^1 in combination with R^2 forms a fused aromatic or heteroaromatic ring that is optionally sulfonated one or more times, or said ring is substituted by -L-R_X or -L-S_C;

R^3 and R^4 are independently selected from the group consisting of [H] hydrogen, C_1-C_6 alkyl, an aromatic or heteroaromatic ring, L-R_X and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, hydroxy, or halogen[; or] and said [an] aromatic or heteroaromatic ring [that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 alkoxy, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_X; or -L-S_C;]

or R^2 in combination with R^3, or R^3 in combination with R^4, forms a 5- or 6-membered alicyclic ring;

R^5 is independently selected from the group consisting of [H] hydrogen, methyl, carboxymethyl, [a] C_2-C_6 alkyl, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen[; or R^5 is an] and said aryl or heteroaryl [ring that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_X; or -L-S_C;]

or R^4 in combination with R^5, or R^5 in combination with R^6, forms a 5- or 6-membered alicyclic ring;

one of X and E is O, S, NR^8, or $CR^{1'}=CR^{2'}$[;], and the other is absent;

wherein R^8 is independently selected from the group consisting of [H] hydrogen, methyl, carboxymethyl, [or a] C_2-C_6 alkyl, -L-R_X and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; [or -L-R_X; or -L-S_C:] and

$R^{1'}$ and $R^{2'}$ are independently selected from the group consisting of [H] hydrogen, cyano, halogen, carboxylic acid, [or] sulfonic acid[; or] , [a] C_1-C_6 alkyl, [or] C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy [that] is optionally substituted by carboxylic acid, sulfonic acid, or halogen[; or an aryl or heteroaryl ring] and said aryl or heteroaryl [that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_X; or -L-S_C;]

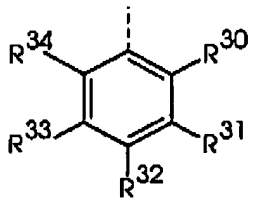
R^{20} and R^{21} are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, [or] sulfonic acid[; or a], C_1-C_6 alkyl [or], C_1-C_6 alkoxy, aromatic or heteroaromatic ring, -L-R_X and -L-S_C, wherein said alkyl or alkoxy [that] is optionally substituted by carboxylic acid, sulfonic acid, or halogen[; or an] said aromatic or heteroaromatic ring [that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_X; or -L-S_C;]

J is O or $NR^{37}R^{38}$;

wherein R^{37} and R^{38} are independently selected from the group consisting of [H] hydrogen, C_1-C_6 alkyl, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen[; an aryl or heteroaryl ring]; or R^{37} in combination with R^{38} forms a saturated 5- or 6-membered heterocycle that is a piperidine, a morpholine, a pyrrolidine or a piperazine, wherein said heterocycle is [each of which is] optionally substituted by methyl, carboxylic acid, or a carboxylic acid ester of a C_1-C_6 alkyl; [or -L-R_x or -L-S_C;]

or R^{37} in combination with R^{20}, or R^{38} in combination with R^{21}, or both, form a 5- or 6-membered ring that is saturated or unsaturated, and is optionally substituted by one or more sulfonic acids, or C_1-C_6 alkyl that is optionally substituted by sulfonic acid;

Q is N or CR^{28}, wherein R^{28} is independently selected from the group consisting of [H] hydrogen, F, CN, carboxylic acid, [or] a carboxylic acid ester of a C_1-C_6 alcohol[; or R^{28} is] , a C_1-C_6 alkyl, -L-R_x and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; or R^{28} [has the] comprises a formula



wherein R^{30}, R^{31}, R^{32}, R^{33} and R^{34} are independently selected from the group consisting of [H] hydrogen, F, Cl, Br, I, sulfonic acid, carboxylic acid, CN, nitro, hydroxy, azido, amino, [hydrazine; or] hydrazino, C_1-C_{18} alkyl, C_1-C_{18} alkoxy, C_1-C_{18} alkylthio, C_1-C_{18} alkanoylamino, C_1-C_{18} alkylaminocarbonyl, C_2-C_{36} dialkylaminocarbonyl, C_1-C_{18} alkyloxycarbonyl, [or C_6-C_{18}] C_7-C_{18} arylcarboxamido, -L-R_x and -L-S_C, wherein said [the] alkyl or aryl portions of said R^{30}, R^{31}, R^{32}, R^{33} and R^{34} [which] are optionally substituted one or more times by substituents selected from the group consisting of F, Cl, Br, I, hydroxy, carboxylic acid, a carboxylic acid ester of a C_1-C_6 alcohol, sulfonic acid, amino, C_1-C_6 alkylamino, C_2-C_6 dialkylamino [or] and C_1-C_6 alkoxy[, the alkyl portions of each having 1-6 carbons]; or [one] a pair of adjacent R^{30}, R^{31}, R^{32}, R^{33} and R^{34} substituents [R^{31} and R^{32}, R^{32} and R^{33} or R^{33} and R^{34},] when taken in combination, form a fused 6-membered aromatic ring that is optionally further substituted by carboxylic acid; [or one or more of R^{30}, R^{31}, R^{32}, R^{33} and R^{34} is -L-R_x or -L-S_C;] and

wherein L is a covalent linkage;

R_x is a reactive group; and

S_C is a conjugated substance.

16. (Amended) [A] The compound[, as claimed in] according to Claim 15, wherein said Q is N.

17. (Amended) [A] The compound[, as claimed in] according to Claim 15, wherein said J is O and said Q is CR^{28}.

18. (Amended) [A] The compound[, as claimed in] according to Claim 17, wherein one of said R^5, R^{21}, R^{30}, R^{31}, R^{32}, R^{33}, and R^{34} is -L-R_x or -L-S_c.

19. (Amended) [A] The compound[, as claimed in] according to Claim 15, wherein

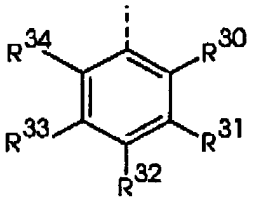
said R^3 and R^4 are each methyl;

R^1 is H or a sulfonic acid;

R^6 is H; and

J is $NR^{37}R^{38}$.

20. (Amended) [A] The compound[, as claimed in] according to Claim 19, wherein Q [has the formula] is CR^{28}[, wherein] and R^{28} has the formula



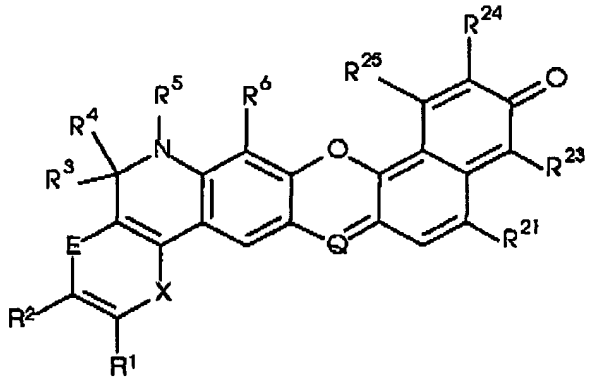
wherein one of R^{30}, R^{31}, R^{32}, R^{33}, and R^{34} is -L-R_x or -L-S_c; and
wherein L is a single covalent bond, or L is a covalent linkage having 1-2[4]0 nonhydrogen atoms selected from the group consisting of C, N, O, P, and S [and is composed of any combination of single, double, triple or aromatic carbon–carbon bonds,

carbon–nitrogen bonds, nitrogen–nitrogen bonds, carbon–oxygen bonds, carbon–sulfur bonds, phosphorus-oxygen bonds, and phosphorus-nitrogen bonds], and

wherein R_X[, when present,] is independently selected from the group consisting of an acrylamide, an activated ester of a carboxylic acid, an acyl azide, an acyl nitrile, an aldehyde, an alkyl halide, an amine, an anhydride, an aniline, an aryl halide, an azide, an aziridine, a boronate, a carboxylic acid, a diazoalkane, a haloacetamide, a halotriazine, a hydrazine, an imido ester, an isocyanate, an isothiocyanate, a maleimide, a phosphoramidite, a reactive platinum complex, a sulfonyl halide, [or] and a thiol group; and

wherein S_C [, when present,] is independently selected from the group consisting of an amino acid, a peptide, a protein, a tyramine, a [monosaccharide, a polysaccharide] carbohydrate, an ion-complexing moiety, a nucleoside, a nucleotide, an oligonucleotide, a nucleic acid, a hapten, a psoralen, a drug, a hormone, a lipid, a lipid assembly, a polymer, a polymeric microparticle, a biological cell, [or] and a virus.

21. (Amended) A compound [of the] comprising a formula



[wherein]

wherein R^1, R^2, and R^6 are independently selected from the group consisting of [H] hydrogen, cyano, halogen, carboxylic acid, [or] sulfonic acid[; or a] C_1-C_6 alkyl, [or] C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy [that] is optionally

substituted by carboxylic acid, sulfonic acid, or halogen[; or an aromatic or heteroaromatic ring that] and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_X; or -L-S_C;]

or R^1 in combination with R^2 forms a fused aromatic or heteroaromatic ring that is optionally sulfonated one or more times, or said ring is substituted by -L-R_X or -L-S_C;

R^3 and R^4 are independently selected from the group consisting of hydrogen, C_1-C_6 alkyl, an aromatic or heteroaromatic ring, L-R_X and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, hydroxy, or halogen[; or] and said [an] aromatic or heteroaromatic ring [that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 alkoxy, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

or R^2 in combination with R^3, or R^3 in combination with R^4, forms a 5- or 6-membered alicyclic ring;

R^5 is independently selected from the group consisting of [H] hydrogen, methyl, carboxymethyl, [a] C_2-C_6 alkyl, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen[; or R^5 is an] and said aryl or heteroaryl [ring that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

or R^4 in combination with R^5, or R^5 in combination with R^6, forms a 5- or 6-membered alicyclic ring;

one of E and X is O, S, NR^8, or $CR^{1'}=CR^{2'}$[;], and the other is absent;

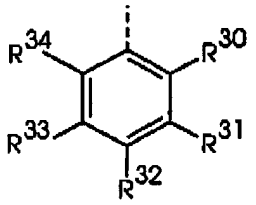
wherein R^8 is independently selected from the group consisting of [H] hydrogen, methyl, carboxymethyl, [or a] C_2-C_6 alkyl, -L-R_X and -L-S_C, wherein said alkyl

[that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; and

$R^{1'}$ and $R^{2'}$ are independently selected from the group consisting of [H] hydrogen, cyano, halogen, carboxylic acid, [or] sulfonic acid[; or] , [a] C_1-C_6 alkyl, [or] C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_x and -L-S_C, wherein said alkyl or alkoxy [that] is optionally substituted by carboxylic acid, sulfonic acid, or halogen[; or an aryl or heteroaryl ring] and said aryl or heteroaryl [that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

R^{21}, R^{23}, R^{24}, and R^{25} are independently selected from the group consisting of hydrogen, cyano, nitro, halogen, carboxylic acid, [or] sulfonic acid[; or a], C_1-C_6 alkyl [or], aromatic or heteroaromatic ring, -L-R_x and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, or halogen[; or an] said aromatic or heteroaromatic ring [that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_x; or -L-S_C;]

Q is N or CR^{28}, wherein R^{28} is independently selected from the group consisting of [H] hydrogen, F, CN, carboxylic acid, [or] a carboxylic acid ester of a C_1-C_6 alcohol[; or R^{28} is] , a C_1-C_6 alkyl, -L-R_x and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; or R^{28} [has the] comprises a formula



wherein R^{30}, R^{31}, R^{32}, R^{33} and R^{34} are independently selected from the group consisting of [H] hydrogen, F, Cl, Br, I, sulfonic acid, carboxylic acid, CN, nitro, hydroxy, azido, amino, hydrazino[; or], C_1-C_{18} alkyl, C_1-C_{18} alkoxy, C_1-C_{18} alkylthio, C_1-C_{18}

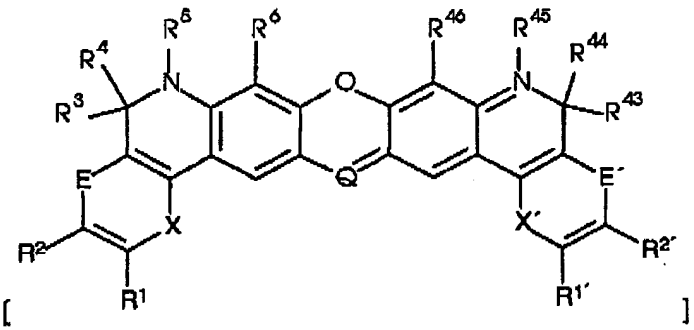
alkanoylamino, C_1-C_{18} alkylaminocarbonyl, C_2-C_{36} dialkylaminocarbonyl, C_1-C_{18} alkyloxycarbonyl, [or C_6-C_{18}] C_7-C_{18} arylcarboxamido, -L-R_x and -L-S_c, wherein said [the] alkyl or aryl portions of said R^{30}, R^{31}, R^{32}, R^{33} and R^{34} [which] are optionally substituted one or more times by substituents selected from the group consisting of F, Cl, Br, I, hydroxy, carboxylic acid, a carboxylic acid ester of a C_1-C_6 alcohol, sulfonic acid, amino, C_1-C_6 alkylamino, C_2-C_6 dialkylamino [or] and C_1-C_6 alkoxy[, the alkyl portions of each having 1-6 carbons]; or [one] a pair of adjacent R^{30}, R^{31}, R^{32}, R^{33} and R^{34} substituents [R^{31} and R^{32}, R^{32} and R^{33} or R^{33} and R^{34},] when taken in combination, form a fused 6-membered aromatic ring that is optionally further substituted by carboxylic acid; [or one or more of R^{30}, R^{31}, R^{32}, R^{33} and R^{34} is -L-R_x or -L-S_c;] and

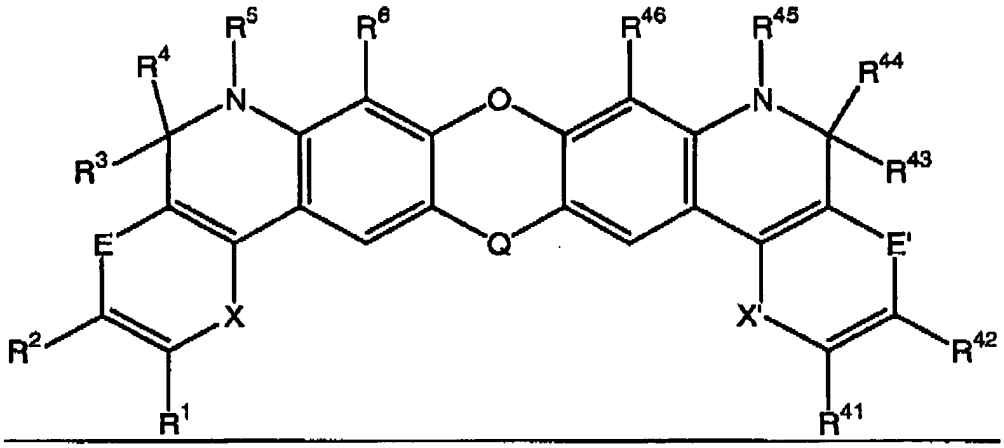
wherein L is a covalent linkage;

R_x is a reactive group; and

S_c is a conjugated substance.

22. (Amended) A compound [of the] comprising a formula:

[]



[wherein]

wherein R^1, R^2, R^6 R^{41}, R^{42}, and R^{46} are independently selected from the group consisting of [H] hydrogen, cyano, [nitro,] halogen, carboxylic acid, [or] sulfonic acid[; or a] C_1-C_6 alkyl, [or] C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy [that] is optionally substituted by carboxylic acid, sulfonic acid, or halogen[; or an aromatic or heteroaromatic ring that] and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_8 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_X; or -L-S_C;]

or R^1 in combination with R^2, or R^{41} in combination with R^{42}, or both, forms a fused aromatic or heteroaromatic ring that is optionally sulfonated one or more times, or said ring is substituted by -L-R_X or -L-S_C;

R^3, R^4, R^{43}, and R^{44} are independently selected from the group consisting of [H] hydrogen, C_1-C_6 alkyl, an aromatic or heteroaromatic ring, L-R_X and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, hydroxy, or halogen[; or] and said [an] aromatic or heteroaromatic ring [that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_8 alkoxy, C_1-C_8 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

or R^2 in combination with R^3, or R^{42} in combination with R^{43}, or R^3 in combination with R^4, or R^{43} in combination with R^{44}, or any combination thereof, forms a 5- or 6-membered alicyclic ring;

R^5 and R^{45} are independently selected from the group consisting of [H] hydrogen, methyl, carboxymethyl, [a] C_2-C_6 alkyl, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen[; or R^5 is an] and said aryl or heteroaryl [ring that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

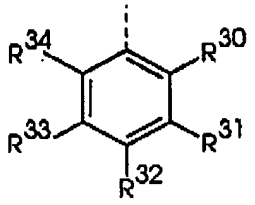
or R^4 in combination with R^5, or R^5 in combination with R^6, or R^{44} in combination with R^{45}, or R^{45} in combination with R^{46}, or any combination thereof, forms a 5- or 6-membered alicyclic ring;

wherein one of said E [and], E', X' and X is O, S, NR^8, or $CR^{1'}=CR^{2'}$[; the other is absent; and one of E′ and X′ is O, S, NR^8, or $CR^{1'}=CR^{2'}$; the other is absent;] provided that E and X or E' and X' are not both present;

wherein R^8 is independently selected from the group consisting of [H] hydrogen, methyl, carboxymethyl, [or a] C_2-C_6 alkyl, -L-R_X and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; and

$R^{1'}$ and $R^{2'}$ are independently selected from the group consisting of [H] hydrogen, cyano, halogen, carboxylic acid, [or] sulfonic acid[; or] , [a] C_1-C_6 alkyl, [or] C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy [that] is optionally substituted by carboxylic acid, sulfonic acid, or halogen[; or an aryl or heteroaryl ring] and said aryl or heteroaryl [that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

Q is N or CR^{28}, wherein R^{28} is independently selected from the group consisting of [H] hydrogen, F, CN, carboxylic acid, [or] a carboxylic acid ester of a C_1-C_6 alcohol[; or R^{28} is] , a C_1-C_6 alkyl, -L-R_x and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; or R^{28} [has the] comprises a formula



wherein R^{30}, R^{31}, R^{32}, R^{33} and R^{34} are independently selected from the group consisting of [H] hydrogen, F, Cl, Br, I, sulfonic acid, carboxylic acid, CN, nitro, hydroxy, azido, amino, hydrazino[; or], C_1-C_{18} alkyl, C_1-C_{18} alkoxy, C_1-C_{18} alkylthio, C_1-C_{18} alkanoylamino, C_1-C_{18} alkylaminocarbonyl, C_2-C_{36} dialkylaminocarbonyl, C_1-C_{18} alkyloxycarbonyl, [or C_6-C_{18}] C_7-C_{18} arylcarboxamido, -L-R_x and -L-S_C, wherein said [the] alkyl or aryl portions of said R^{30}, R^{31}, R^{32}, R^{33} and R^{34} [which] are optionally substituted one or more times by substituents selected from the group consisting of F, Cl, Br, I, hydroxy, carboxylic acid, a carboxylic acid ester of a C_1-C_6 alcohol, sulfonic acid, amino, C_1-C_6 alkylamino, C_2-C_6 dialkylamino [or] and C_1-C_6 alkoxy[, the alkyl portions of each having 1-6 carbons]; or [one] a pair of adjacent R^{30}, R^{31}, R^{32}, R^{33} and R^{34} substituents [R^{31} and R^{32}, R^{32} and R^{33} or R^{33} and R^{34},] when taken in combination, form a fused 6-membered aromatic ring that is optionally further substituted by carboxylic acid; [or one or more of R^{30}, R^{31}, R^{32}, R^{33} and R^{34} is -L-R_x or -L-S_C;] and

wherein L is a covalent linkage;

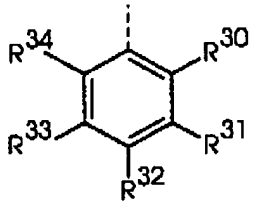
R_x is a reactive group; and

S_C is a conjugated substance.

23. (Cancel) A compound, as claimed in Claim 22, wherein

$X = X'$, $E = E'$, $R^1 = R^{41}$, and $R^2 = R^{42}$.

24. (Amended) [A] The compound[, as claimed in] according to Claim 22, wherein Q [has the formula] is CR^{28}[, wherein] and R^{28} has the formula



25. (Amended) [A] The compound[, as claimed in] according to Claim 24, wherein one of R^5, [R^{21},] R^{30}, R^{31}, R^{32}, R^{33}, R^{34}, and R^{45} is -L-R_x or -L-S_c.

26. (Amended) [A] The compound[, as claimed in] according to Claim 24, wherein

said R^3, R^4, R^{43}, and R^{44} are each methyl;

each R^1 and R^{41} [are] is independently H or sulfonic acid; and

R^6 and R^{46} are H.

27. (Amended) [A] The compound[, as claimed in] according to Claim 24, wherein [the] said compound is substituted one or more times by sulfonic acid.

28. (Amended) [A] The compound[, as claimed in] according to Claim 22, wherein one of said R^1, $R^{1'}$, R^2, $R^{2'}$, R^3, R^4, R^5, R^6, [R^7, R^8, R^{15}, R^{16}, R^{20}, R^{21}, R^{23}, R^{24}, R^{25},] R^{28}, R^{30}, R^{31}, R^{32}, R^{33}, R^{34}, [R^{37}, R^{38},] R^{41}, R^{42}, R^{43}, R^{44}, R^{45}, and R^{46} is [an] -L-R_x or -L-S_c.

29. (Amended) [A] The compound[, as claimed in] according to Claim 28, wherein each L is independently a single covalent bond, or L is a covalent linkage having 1-2[4]0

nonhydrogen atoms selected from the group consisting of C, N, O, P, and S. [and is composed of any combination of single, double, triple or aromatic carbon–carbon bonds, carbon–nitrogen bonds, nitrogen–nitrogen bonds, carbon–oxygen bonds, carbon–sulfur bonds, phosphorus-oxygen bonds, and phosphorus-nitrogen bonds.]

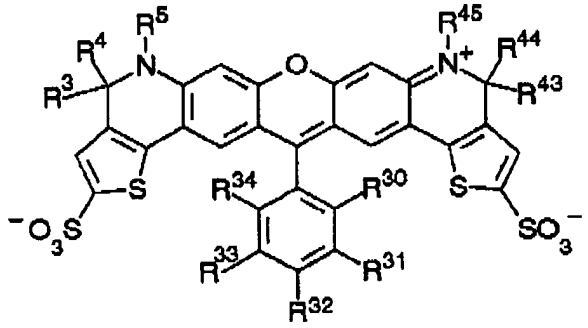
30. (Amended) [A] The compound[, as claimed in] according to Claim 28, wherein said R_X is independently selected from the group consisting of an acrylamide, an activated ester of a carboxylic acid, an acyl azide, an acyl nitrile, an aldehyde, an alkyl halide, an amine, an anhydride, an aniline, an aryl halide, an azide, an aziridine, a boronate, a carboxylic acid, a diazoalkane, a haloacetamide, a halotriazine, a hydrazine, an imido ester, an isocyanate, an isothiocyanate, a maleimide, a phosphoramidite, a reactive platinum complex, a sulfonyl halide, a perfluorobenzamido, an azidoperfluorobenzamido group, [or] and a thiol group.

31. (Amended) [A] The compound[, as claimed in] according to Claim [28] 30, wherein said R_X is independently selected from the group consisting of a phosphoramidite, a succinimidyl ester of a carboxylic acid, a haloacetamide, a hydrazine, an isothiocyanate, a maleimide group, a perfluorobenzamido, an azidoperfluorobenzamido group, [or] and a reactive platinum complex.

32. (Amended) [A] The compound[, as claimed in] according to Claim 28, wherein said S_O is independently selected from the group consisting of an amino acid, a peptide, a protein, a tyramine, a [monosaccharide, a polysaccharide] carbohydrate, an ion-complexing moiety, a nucleoside, a nucleotide, an oligonucleotide, a nucleic acid, a hapten, a psoralen, a drug, a hormone, a lipid, a lipid assembly, a polymer, a polymeric microparticle, a biological cell, [or] and a virus.

33. (Amended) [A] The compound[, as claimed in] according to Claim [28] 32, wherein S_C is an amino acid, a peptide, a protein, an ion-complexing moiety, a nucleoside, a nucleotide, an oligonucleotide, [or] and a nucleic acid.

34. (Amended) [A] The compound[, as claimed in] according to Claim 28, wherein said compound comprises a [having the] formula:



wherein said R^3, R^4, R^5, R^{43}, R^{44} , and R^{45} are independently methyl or ethyl;
R^{30} is sulfonic acid or carboxylic acid;
R^{31} and R^{34} are independently H, F, or Cl;
one of R^{32} and R^{33} is H, F, or Cl, and the other of R^{32} and R^{33} is -L-R_x or -L-S_c,

wherein said L is a covalent linkage [of the formula] comprising –
$S(CH_2)_aCOO(CH_2)_b$— or [the formula] –$S(CH_2)_aCONH(CH_2)_b$—
wherein a is an integer between 0 and 10, and b is an integer between 0 and 10 [provided that a and b are not both 0]; and
wherein said R_x [, where present,] is selected from the group consisting of a carboxylic acid, an activated ester of a carboxylic acid, a haloacetamide, a hydrazine, an isothiocyanate, a maleimide group, [or] and a reactive platinum complex.; and
wherein said S_c[, where present,] is selected from the group consisting of an amino acid, a peptide, a protein, an ion-complexing moiety, a nucleoside, a nucleotide, an oligonucleotide, a lectin, or a nucleic acid.

35. (Amended) [A] The compound[, as claimed in] according to Claim 34, wherein said R_x is a maleimide group or is a succinimidyl ester of a carboxylic acid.

36. (Amended) [A] The compound[, as claimed in] according to Claim 34, wherein said S_c is a peptide or a protein [or a lectin].

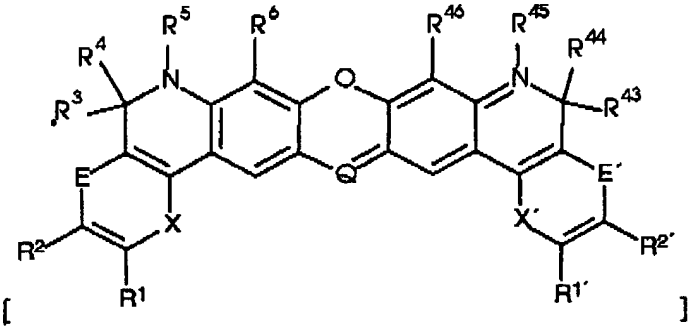
37. (Amended) [A] The compound[, as claimed in] according to Claim 3[4]6, wherein said S_c is an antibody or antibody fragment or a lectin.

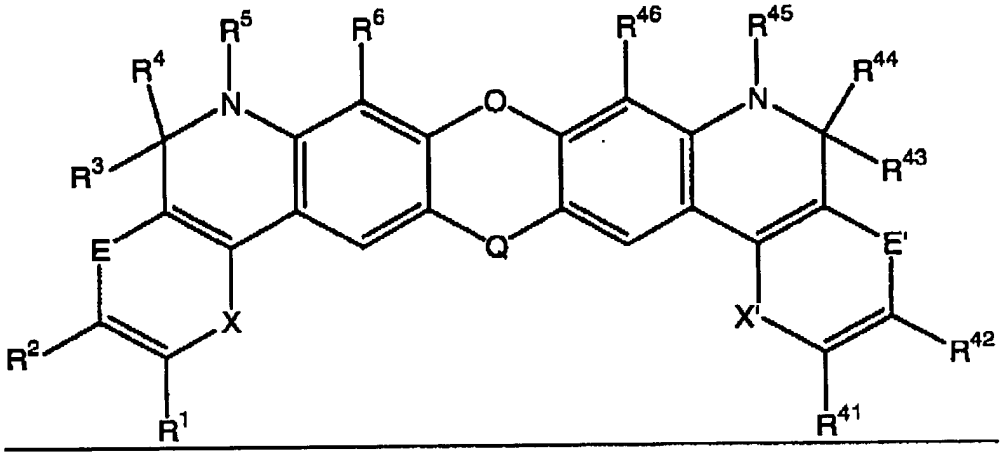
38. (Amended) [A] The compound[, as claimed in] according to Claim 34, wherein said S_c is a nucleotide or an oligonucleotide.

39. (Amended) [A] The compound[, as claimed in] according to Claim 34, wherein said S_c is a BAPTA or APTRA ion-complexing moiety.

40. (Amended) A method of staining a [biological] sample, said method comprising steps:

a) combining a [dye] solution with said sample, wherein said solution comprises [comprising] a compound [of the] having formula

[]



[wherein]

wherein R^1, R^2, R^6, R^{41}, R^{42}, and R^{46} are independently selected from the group consisting of [H] hydrogen, cyano, [nitro,] halogen, carboxylic acid, [or] sulfonic acid[; or a] C_1-C_6 alkyl, [or] C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_x and -L-S_c, wherein said alkyl or alkoxy [that] is optionally substituted by carboxylic acid, sulfonic acid, or halogen[; or an aromatic or heteroaromatic ring that] and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl; [or -L-R_x; or -L-S_c;]

or R^1 in combination with R^2, or R^{41} in combination with R^{42}, or both, forms a fused aromatic or heteroaromatic ring that is optionally sulfonated one or more times, or said ring is substituted by -L-R_x or -L-S_c;

R^3, R^4, R^{43}, and R^{44} are independently selected from the group consisting of [H] hydrogen, C_1-C_6 alkyl, an aromatic or heteroaromatic ring, L-R_x and -L-S_c, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, hydroxy, or halogen[; or] and said [an] aromatic or heteroaromatic ring [that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 alkoxy, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

or R^2 in combination with R^3, or R^{42} in combination with R^{43}, or R^3 in combination with R^4, or R^{43} in combination with R^{44}, or any combination thereof, forms a 5- or 6-membered alicyclic ring;

R^5 and R^{45} are independently selected from the group consisting of [H] hydrogen, methyl, carboxymethyl, [a] C_2-C_6 alkyl, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen[; or R^5 is an] and said aryl or heteroaryl [ring that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

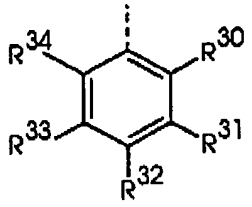
or R^4 in combination with R^5, or R^5 in combination with R^6, or R^{44} in combination with R^{45}, or R^{45} in combination with R^{46}, or any combination thereof, forms a 5- or 6-membered alicyclic ring;

wherein one of said E [and], E', X' and X is O, S, NR^8, or $CR^{1'}=CR^{2'}$[; the other is absent; and one of E' and X' is O, S, NR^8, or $CR^{1'}=CR^{2'}$; the other is absent;] provided that E and X or E' and X' are not both present;

wherein R^8 is independently selected from the group consisting of [H] hydrogen, methyl, carboxymethyl, [or a] C_2-C_6 alkyl, -L-R_X and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; and

$R^{1'}$ and $R^{2'}$ are independently selected from the group consisting of [H] hydrogen, cyano, halogen, carboxylic acid, [or] sulfonic acid[; or], [a] C_1-C_6 alkyl, [or] C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy [that] is optionally substituted by carboxylic acid, sulfonic acid, or halogen[; or an aryl or heteroaryl ring] and said aryl or heteroaryl [that] is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

Q is N or CR^{28}, wherein R^{28} is independently selected from the group consisting of [H] hydrogen, F, CN, carboxylic acid, [or] a carboxylic acid ester of a C_1-C_6 alcohol[; or R^{28} is] , a C_1-C_6 alkyl, -L-R_x and -L-S_C, wherein said alkyl [that] is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; or R^{28} [has the] comprises a formula



wherein R^{30}, R^{31}, R^{32}, R^{33} and R^{34} are independently selected from the group consisting of [H] hydrogen, F, Cl, Br, I, sulfonic acid, carboxylic acid, CN, nitro, hydroxy, azido, amino, hydrazino[; or], C_1-C_{18} alkyl, C_1-C_{18} alkoxy, C_1-C_{18} alkylthio, C_1-C_{18} alkanoylamino, C_1-C_{18} alkylaminocarbonyl, C_2-C_{36} dialkylaminocarbonyl, C_1-C_{18} alkyloxycarbonyl, [or C_6-C_{18}] C_7-C_{18} arylcarboxamido, -L-R_x and -L-S_C, wherein said [the] alkyl or aryl portions of said R^{30}, R^{31}, R^{32}, R^{33} and R^{34} [which] are optionally substituted one or more times by substituents selected from the group consisting of F, Cl, Br, I, hydroxy, carboxylic acid, a carboxylic acid ester of a C_1-C_6 alcohol, sulfonic acid, amino, C_1-C_6 alkylamino, C_2-C_6 dialkylamino [or] and C_1-C_6 alkoxy[, the alkyl portions of each having 1-6 carbons]; or [one] a pair of adjacent R^{30}, R^{31}, R^{32}, R^{33} and R^{34} substituents [R^{31} and R^{32}, R^{32} and R^{33} or R^{33} and R^{34},] when taken in combination, form a fused 6-membered aromatic ring that is optionally further substituted by carboxylic acid; [or one or more of R^{30}, R^{31}, R^{32}, R^{33} and R^{34} is -L-R_x or -L-S_C;] and

wherein L is a covalent linkage;

R_x is a reactive group; and

S_C is a conjugated substance;

[with a biological sample in a concentration sufficient to yield a detectable optical response under the desired conditions.]

b) illuminating said sample with a suitable light wavelength to yield a detectable optical response.

41. (Amended) [A] The method [, as claimed in] according to Claim 40, wherein said method further [comprising] comprises combining [the] said sample with an additional detection reagent [that has spectral properties that are detectably different from said optical response].

42. (Cancelled) A method, as claimed in Claim 40, further comprising the step of determining a characteristic of the sample by comparing the optical response with a standard response parameter.

43. (Amended) [A] The method [, as claimed in] according to Claim 40, wherein [the] said sample comprises cells, growth medium, tissue, proteins, peptides, or biological fluids.

44. (Amended) [A] The method [, as claimed in] according to Claim 40, wherein [the] said sample is immobilized in or on a solid or semi-solid matrix that is a membrane, an electrophoretic gel, a silicon chip, a glass slide, a microwell plate, or a microfluidic chip.

45. (Cancelled) A method, as claimed in Claim 40, further comprising tracing the temporal or spatial location of the optical response within the sample.

46. (Amended) [A] The method[, as claimed in] according to Claim 40, wherein [for said compound] at least one of said R^{28}, R^{30}, R^{31}, R^{32}, R^{33}, and R^{34}[, R^{37} and R^{38}] is -L-R_x or -L-S_C;

R_x is selected from the group consisting of a carboxylic acid, an activated ester of a carboxylic acid, an amine, an azide, a hydrazine, a haloacetamide, an alkyl

halide, an isothiocyanate, [or] and a maleimide group; and

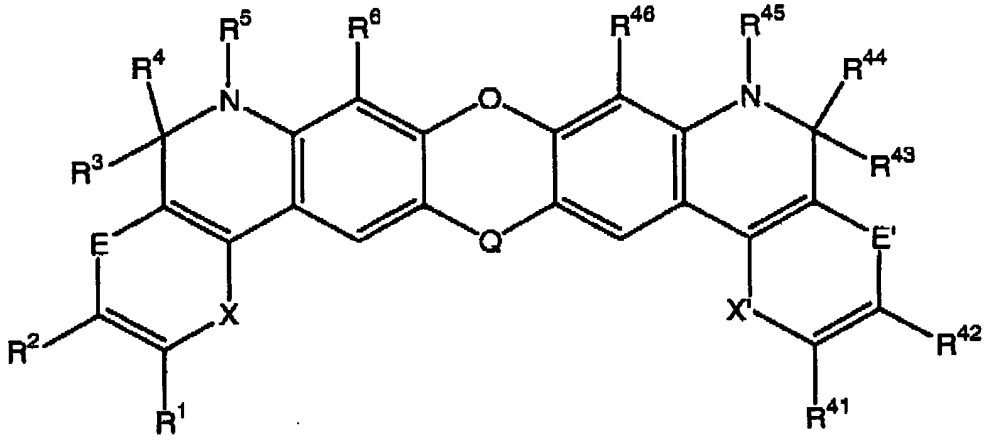
S_c is selected from the group consisting of an amino acid, a peptide, a protein, a polysaccharide, a nucleotide, a nucleoside, an oligonucleotide, a nucleic acid polymer, an ion-complexing moiety, a lipid, or a non-biological organic polymer or polymeric microparticle, wherein said Sc [that] is optionally bound to one or more additional fluorophores [that are the same or different].

47. (Amended) [A] The method[, as claimed in] according to Claim 46, wherein [for said compound,] said R^{28} is an -L-S_c, and S_c is an ion-complexing moiety that is a BAPTA or an APTRA.

48. (Amended) [A] The method[, as claimed in] according to Claim 4[0]6, wherein at least one of said R^{28}, R^{30}, R^{31}, R^{32}, R^{33}, and R^{34}, [R^{37} and R^{38}] is -L-S_c, and said S_c is a nucleoside, a nucleotide, an oligonucleotide, or a nucleic acid polymer.

49. (New) A kit for staining a sample, wherein said kit comprises

a solution comprising a buffer and a compound having formula



wherein R^1, R^2, R^6, R^{41}, R^{42} and R^{46} are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_x and -L-S_c, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

or R^1 in combination with R^2, or R^{41} in combination with R^{42}, or both, forms a fused aromatic or heteroaromatic ring that is optionally sulfonated one or more times, or said ring is substituted by -L-R_x or -L-S_c;

R^3, R^4, R^{43}, and R^{44} are independently selected from the group consisting of hydrogen, C_1-C_6 alkyl, an aromatic ring, a heteroaromatic ring, L-R_x and -L-S_c, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, hydroxy, or halogen and said aromatic or heteroaromatic ring is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 alkoxy, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

or R^2 in combination with R^3, or R^{42} in combination with R^{43}, or R^3 in combination with R^4, or R^{43} in combination with R^{44}, or any combination thereof, forms a 5- or 6-membered alicyclic ring;

R^5 and R^{45} are independently selected from the group consisting of hydrogen, methyl, carboxymethyl, C_2-C_6 alkyl, aryl, heteroaryl, -L-R_x and -L-S_c, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

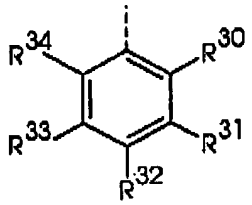
or R^4 in combination with R^5, or R^5 in combination with R^6, or R^{44} in combination with R^{45}, or R^{45} in combination with R^{46}, or any combination thereof, forms a 5- or 6-membered alicyclic ring;

wherein one of said E, E', X' and X is O, S, NR^8, or $CR^{1'}=CR^{2'}$, provided that E and X or E' and X' are not both present;

wherein R^8 is independently selected from the group consisting of hydrogen, methyl, carboxymethyl, C_2-C_6 alkyl, -L-R_X and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; and

$R^{1'}$ and $R^{2'}$ are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid, C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

Q is N or CR^{28}, wherein R^{28} is independently selected from the group consisting of hydrogen, F, CN, carboxylic acid, a carboxylic acid ester of a C_1-C_6 alcohol, a C_1-C_6 alkyl, -L-R_X and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; or R^{28} comprises a formula



wherein R^{30}, R^{31}, R^{32}, R^{33} and R^{34} are independently selected from the group consisting of hydrogen, F, Cl, Br, I, sulfonic acid, carboxylic acid, CN, nitro, hydroxy, azido, amino, hydrazine, C_1-C_{18} alkyl, C_1-C_{18} alkoxy, C_1-C_{18} alkylthio, C_1-C_{18} alkanoylamino, C_1-C_{18} alkylaminocarbonyl, C_2-C_{36} dialkylaminocarbonyl, C_1-C_{18} alkyloxycarbonyl, C_7-C_{18} arylcarboxamido, -L-R_x and -L-S_C, wherein said alkyl or aryl portions of said R^{30}, R^{31}, R^{32}, R^{33} and R^{34} are optionally substituted one or more times by substituents selected from

the group consisting of F, Cl, Br, I, hydroxy, carboxylic acid, a carboxylic acid ester of a C_1-C_6 alcohol, sulfonic acid, amino, C_1-C_6 alkylamino, C_2-C_6 dialkylamino and C_1-C_6 alkoxy; or a pair of adjacent R^{30}, R^{31}, R^{32}, R^{33} and R^{34} substituents when taken in combination, form a fused 6-membered aromatic ring that is optionally further substituted by carboxylic acid; and

wherein L is a covalent linkage;

R_x is a reactive group; and

S_c is a conjugated substance.

50. (New) The kit according to Claim 49, wherein said kit further comprises an additional detection reagent, a purification medium, or standards.

51. (New) The kit according to Claim 49, wherein at least one of said R^1, R^2, R^3, R^4, R^5, R^6, R^{28}, R^{30}, R^{31}, R^{32}, R^{33}, R^{34}, R^{43}, R^{44}, R^{45} and R^{46} is L-R_x wherein said R_x is independently selected from the group consisting of a carboxylic acid, an activated ester of a carboxylic acid, an amine, an azide, a hydrazine, a haloacetamide, an alkyl halide, an isothiocyanate, and a maleimide group.

52. (New) The kit according to Claim 51, wherein at least one of said R^{31}, R^{32}, R^{33}, or R^{34} is L-R_x and R^{30} is carboxylic acid or sulfonic acid.

53. (New) The kit according to Claim 49, wherein at least one of said R^1, R^2, R^3, R^4, R^5, R^6, R^{28}, R^{30}, R^{31}, R^{32}, R^{33}, R^{34}, R^{43}, R^{44}, R^{45} and R^{46} is L-Sc, wherein said Sc is independently selected from the group consisting of an amino acid, a peptide, a protein, an antibody, an antibody fragment, a carbohydrate, a nucleotide, a nucleoside, an oligonucleotide, a nucleic acid polymer, an ion-complexing moiety, a lipid, a non-biological organic polymer and polymeric microparticle.

54. (New) The kit according to Claim 53, wherein said Sc is an antibody or fragment thereof.

Respectfully submitted,

Date: December 19, 2002

Koren J. Anderson

Koren J. Anderson, PhD
Reg. No. 51,061

Molecular Probes, Inc.
P.O. Box 22010
Eugene, Oregon, 97402
Phone: (541) 984-5656
Facsimile: (541) 465- 8354

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re application of: Diwu *et al.*	
Serial No.: 09/922,333	Examiner: F. Powers
Filed: August 4, 2001	Group Art Unit: 1626
For: **DERIVATIVES OF 1,2-DIHYDRO-7-HYDROXYQUINOLINES CONTAINING FUSED RINGS**	**CLEAN VERSION OF THE CLAIMS**

Assistant Commissioner for Patents
U.S. Patent and Trademark Office
Washington, D.C. 20231

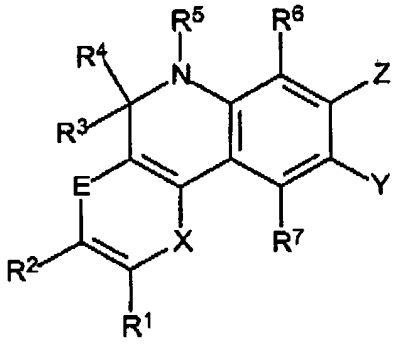
Dear Sir:

The following Marked-up Version of the Claims is hereby submitted together with a Clean Version of the Claims and the Response to Notice of Non-Compliant Amendment (37 CFR 1.121) on or before the due date of December 25, 2002.

CERTIFICATE OF TRANSMISSION

I HEREBY CERTIFY THAT THIS PAPER AND THE DOCUMENTS REFERRED AS BEING ATTACHED OR ENCLOSED HEREWITH ARE BEING FACSIMILE TRANSMITTED TO THE UNITED STATES PATENT AND TRADMARK OFFICE ON 12/19/02 TO 1.703.872.9306 By Judith M. Ubando

1. (Amended) A compound comprising a formula



wherein

R^1 and R^2 are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid, C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

or R^1 in combination with R^2 forms a fused aromatic or heteroaromatic ring that is optionally sulfonated one or more times, or said ring is substituted by -L-R_X or -L-S_C;

or R^2 in combination with R^3 forms a 5- or 6-membered alicyclic ring;

R^3 and R^4 are independently selected from the group consisting of hydrogen, C_1-C_6 alkyl, aromatic or heteroaromatic ring, -L-R_X and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, hydroxy, or halogen and said aromatic or heteroaromatic ring is optionally substituted one or more times by C_1-C_6

alkyl, C_1-C_6 alkoxy, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

or R^3 in combination with R^4 forms a 5- or 6-membered alicyclic ring;

R^5 is independently selected from the group consisting of hydrogen, methyl, carboxymethyl, C_2-C_6 alkyl, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

R' Cont.

R^6 is independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid , C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L--S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

or R^4 in combination with R^5, or R^5 in combination with R^6, forms a 5- or 6-membered alicyclic ring;

R^7 is independently selected from the group consisting of hydrogen, C_1-C_6 alkyl, C_1-C_6 alkoxy, -L-R_X and -L-S_C;

one of X and E is O, S, NR^8, or $CR^{1'}$=$CR^{2'}$, and the other is absent;

wherein R^8 is independently selected from the group consisting of hydrogen, methyl, carboxymethyl, C_2-C_6 alkyl, -L-R_X and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; and

$R^{1'}$ and $R^{2'}$ are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid, C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl,

heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_8 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

Y is independently selected from the group consisting of H, OH, NH_2, NO, -(CO)-R^9, -(CO)-O-R^{10}, wherein said R^9 and R^{10} are independently H, C_1-C_6 alkyl, or a substituted or unsubstituted aryl or heteroaryl ring system having 1-2 rings;

Z is independently selected from the group consisting of H, OH, NHR^{17}, SH, or $C(CR^{11}R^{12})_2OH$; wherein said R^{17} is a C_1-C_6 alkyl that is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen and said R^{11} and R^{12} are independently C_1-C_6 alkyl that are optionally substituted by carboxylic acid, sulfonic acid, or halogen, or R^{11} and R^{12} taken in combination form a 5- or 6-membered alicyclic ring;

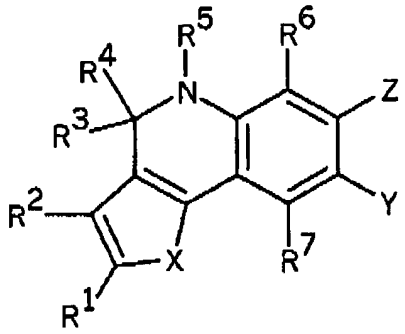
wherein L is a covalent linkage;

R_x is a reactive group; and

S_C is a conjugated substance.

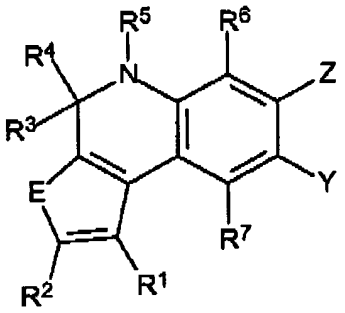
2. (Amended) The compound according to Claim 1, wherein one of X and E is O, S, or $CR^{1'}=CR^{2'}$, and the other is absent.

3. (Amended) The compound according to Claim 2, wherein said compound has the formula



wherein X is O or S.

4. (Amended) The compound according to Claim 2, wherein said compound has the formula



wherein E is O or S.

5. (Amended) The compound according to Claim 3, wherein X is S.

6. (Amended) The compound according to Claim 1, wherein

R^1 is hydrogen or sulfonic acid;

R^3 and R^4 are each methyl;

R^6 and R^7 are each hydrogen or methyl; and

Z is OH.

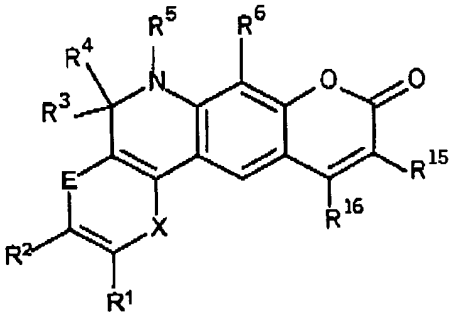
7. (Amended) The compound according to Claim 1, wherein Y is H or -(CO)-H or NO.

8. (Amended) The compound according to Claim 1, wherein said L is independently a single covalent bond or a covalent linkage having 1-20 nonhydrogen atoms selected from the group consisting of C, N, O, P, and S.

9. (Amended) The compound according to Claim 1, wherein said R_X is independently selected from the group consisting of an acrylamide, an activated ester of a carboxylic acid, an acyl azide, an acyl nitrile, an aldehyde, an alkyl halide, an amine, an anhydride, an aniline, an aryl halide, an azide, an aziridine, a boronate, a carboxylic acid, a diazoalkane, a haloacetamide, a halotriazine, a hydrazine, an imido ester, an isocyanate, an isothiocyanate, a maleimide, a phosphoramidite, a reactive platinum complex, a sulfonyl halide, and a thiol group.

10. (Amended) The compound according to Claim 1, wherein said S_C is independently selected from the group consisting of an amino acid, a peptide, a protein, a tyramine, a carbohydrate, an ion-complexing moiety, a nucleoside, a nucleotide, an oligonucleotide, a nucleic acid, a hapten, a psoralen, a drug, a hormone, a lipid, a lipid assembly, a polymer, a polymeric microparticle, a biological cell, and a virus.

11. (Amended) A compound comprising a formula



wherein R^1, R^2, and R^6 are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_x and -L-S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

or R^1 in combination with R^2 forms a fused aromatic or heteroaromatic ring that is optionally sulfonated one or more times, or said ring is substituted by -L-R_X or -L-S_C;

R^3 and R^4 are independently selected from the group consisting of hydrogen, C_1-C_6 alkyl, an aromatic or heteroaromatic ring, L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, hydroxy, or halogen and said aromatic or heteroaromatic ring is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 alkoxy, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

or R^2 in combination with R^3, or R^3 in combination with R^4, forms a 5- or 6-membered alicyclic ring;

R^5 is independently selected from the group consisting of hydrogen, methyl, carboxymethyl, C_2-C_6 alkyl, aryl, heteroaryl, -L-R_x and -L-S_C, wherein said alkyl is

optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

or R^4 in combination with R^5, or R^5 in combination with R^6, forms a 5- or 6-membered alicyclic ring;

one of X and E is O, S, NR^8, or $CR^{1'}=CR^{2'}$ and the other is absent;

wherein R^8 is independently selected from the group consisting of hydrogen, methyl, carboxymethyl, C_2-C_6 alkyl, -L-R_X and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; and

$R^{1'}$ and $R^{2'}$ are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid, C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

R^{15} and R^{16} are independently selected from the group consisting of hydrogen, cyano, nitro, halogen, carboxylic acid, sulfonic acid, C_1-C_6 alkyl, an aromatic or heteroaromatic ring system having 1-2 fused rings, -L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aromatic or heteroaromatic ring system is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

wherein L is a covalent linkage;

R_x is a reactive group; and

S_C is a conjugated substance.

12. (Amended) The compound according to Claim 11, wherein said one of X and E is O or S.

13. (Amended) The compound according to Claim 12, wherein

R^6 and R^7 are hydrogen;

R^3 and R^4 are each methyl;

R^1 is hydrogen or sulfonic acid;

R1 cont.

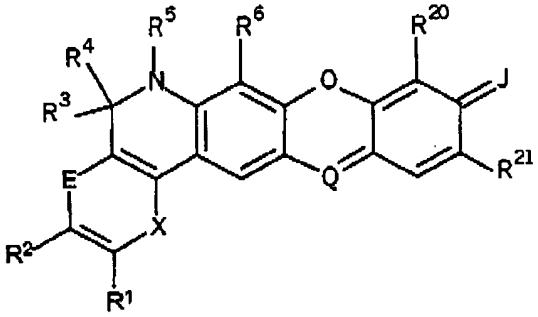
one of R^{15} and R^{16} is -L-R_x or -L-S_C, and the other is hydrogen, C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl; or cyano;

wherein L is a single covalent bond, or L is a covalent linkage having 1-20 nonhydrogen atoms selected from the group consisting of C, N, O, P, and S, and
wherein R_X is independently selected from the group consisting of an acrylamide, an activated ester of a carboxylic acid, an acyl azide, an acyl nitrile, an aldehyde, an alkyl halide, an amine, an anhydride, an aniline, an aryl halide, an azide, an aziridine, a boronate, a carboxylic acid, a diazoalkane, a haloacetamide, a halotriazine, a hydrazine, an imido ester, an isocyanate, an isothiocyanate, a maleimide, a phosphoramidite, a reactive platinum complex, a sulfonyl halide, and a thiol group; and
wherein S_C is independently selected from the group consisting of an amino acid, a peptide, a protein, a tyramine, a carbohydrate, an ion-complexing moiety, a nucleoside, a nucleotide, an oligonucleotide, a nucleic acid, a hapten, a psoralen, a drug, a hormone, a lipid, a lipid assembly, a polymer, a polymeric microparticle, a biological cell, and a virus.

14. (Amended) The compound according to Claim 11, wherein one of said R^{15} or R^{16} is an aromatic or heteroaromatic ring system having 1-2 fused rings that is optionally

substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl.

15. A compound comprising a formula:



wherein R^1, R^2, and R^6 are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_x and -L-S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

or R^1 in combination with R^2 forms a fused aromatic or heteroaromatic ring that is optionally sulfonated one or more times, or said ring is substituted by -L-R_X or -L-S_C;

R^3 and R^4 are independently selected from the group consisting of hydrogen, C_1-C_6 alkyl, an aromatic or heteroaromatic ring, L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, hydroxy, or halogen and said aromatic or heteroaromatic ring is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 alkoxy, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

or R^2 in combination with R^3, or R^3 in combination with R^4, forms a 5- or 6-membered alicyclic ring;

R^5 is independently selected from the group consisting of hydrogen, methyl, carboxymethyl, C_2-C_6 alkyl, aryl, heteroaryl, -L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

or R^4 in combination with R^5, or R^6 in combination with R^6, forms a 5- or 6-membered alicyclic ring;

one of X and E is O, S, NR^8, or $CR^{1'}=CR^{2'}$, and the other is absent;

wherein R^8 is independently selected from the group consisting of hydrogen, methyl, carboxymethyl, C_2-C_6 alkyl, -L-R_X and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; and

$R^{1'}$ and $R^{2'}$ are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid, C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

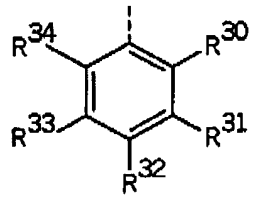
R^{20} and R^{21} are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid, C_1-C_6 alkyl, C_1-C_6, alkoxy, aromatic or heteroaromatic ring, -L-R_x and -L-S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen said aromatic or heteroaromatic ring is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

J is O or $NR^{37}R^{38}$;

wherein R^{37} and R^{38} are independently selected from the group consisting of hydrogen, C_1-C_6 alkyl, aryl, heteroaryl, -L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; or R^{37} in combination with R^{38} forms a saturated 5- or 6-membered heterocycle that is a piperidine, a morpholine, a pyrrolidine or a piperazine, wherein said heterocycle is optionally substituted by methyl, carboxylic acid, or a carboxylic acid ester of a C_1-C_6 alkyl;

or R^{37} in combination with R^{20}, or R^{38} in combination with R^{21}, or both, form a 5- or 6-membered ring that is saturated or unsaturated, and is optionally substituted by one or more sulfonic acids, or C_1-C_6 alkyl that is optionally substituted by sulfonic acid;

Q is N or CR^{28}, wherein R^{28} is independently selected from the group consisting of hydrogen, F, CN, carboxylic acid, a carboxylic acid ester of a C_1-C_6 alcohol, a C_1-C_6 alkyl, -L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; or R^{28} comprises a formula



wherein R^{30}, R^{31}, R^{32}, R^{33} and R^{34} are independently selected from the group consisting of hydrogen, F, Cl, Br, I, sulfonic acid, carboxylic acid, CN, nitro, hydroxy, azido, amino, hydrazino, C_1-C_{18} alkyl, C_1-C_{18} alkoxy, C_1-C_{18} alkylthio, C_1-C_{18} alkanoylamino, C_1-C_{18} alkylaminocarbonyl, C_2-C_{36} dialkylaminocarbonyl, C_1-C_{18} alkyloxycarbonyl, C_7-C_{18} arylcarboxamido, -L-R_x and -L-S_C, wherein said alkyl or aryl portions of said R^{30}, R^{31}, R^{32}, R^{33} and R^{34} are optionally substituted one or more times by substituents selected from

the group consisting of F, Cl, Br, I, hydroxy, carboxylic acid, a carboxylic acid ester of a C_1-C_6 alcohol, sulfonic acid, amino, C_1-C_6 alkylamino, C_2-C_6 dialkylamino and C_1-C_6 alkoxy; or a pair of adjacent R^{30}, R^{31}, R^{32}, R^{33} and R^{34} substituents when taken in combination, form a fused 6-membered aromatic ring that is optionally further substituted by carboxylic acid; and

wherein L is a covalent linkage;

R_x is a reactive group; and

S_C is a conjugated substance.

R2

16. (Amended) The compound according to Claim 15, wherein said Q is N.

17. (Amended) The compound according to Claim 15, wherein said J is O and said Q is CR^{28}.

18. (Amended) The compound according to Claim 17, wherein one of said R^5, R^{21}, R^{30}, R^{31}, R^{32}, R^{33}, and R^{34} is -L-R_x or -L-S_C.

19. (Amended) The compound according to Claim 15, wherein

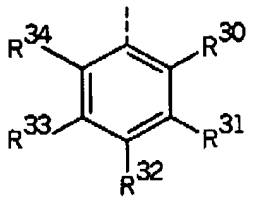
said R^3 and R^4 are each methyl;

R^1 is H or a sulfonic acid;

R^6 is H; and

J is $NR^{37}R^{38}$.

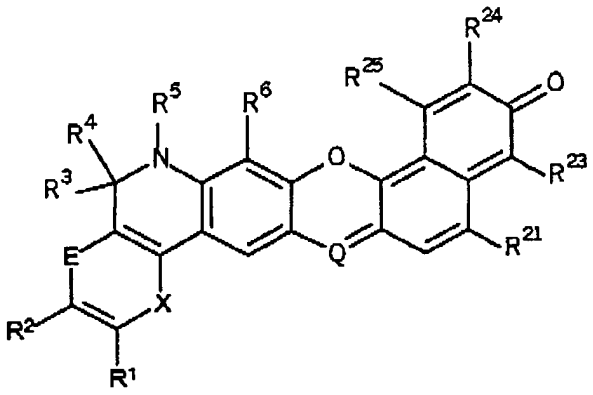
20. (Amended) The compound according to Claim 19, wherein Q is CR^{28} and R^{28} has the formula



wherein one of R^{30}, R^{31}, R^{32}, R^{33}, and R^{34} is -L-R_x or -L-S_C; and
wherein L is a single covalent bond, or L is a covalent linkage having 1-20 nonhydrogen atoms selected from the group consisting of C, N, O, P, and S, and
wherein R_X is independently selected from the group consisting of an acrylamide, an activated ester of a carboxylic acid, an acyl azide, an acyl nitrile, an aldehyde, an alkyl halide, an amine, an anhydride, an aniline, an aryl halide, an azide, an aziridine, a boronate, a carboxylic acid, a diazoalkane, a haloacetamide, a halotriazine, a hydrazine, an imido ester, an isocyanate, an isothiocyanate, a maleimide, a phosphoramidite, a reactive platinum complex, a sulfonyl halide, and a thiol group; and
wherein S_C is independently selected from the group consisting of an amino acid, a peptide, a protein, a tyramine, a carbohydrate, an ion-complexing moiety, a nucleoside, a nucleotide, an oligonucleotide, a nucleic acid, a hapten, a psoralen, a drug, a hormone, a lipid, a lipid assembly, a polymer, a polymeric microparticle, a biological cell, and a virus.

R2 cont.

21. (Amended) A compound comprising a formula



wherein R^1, R^2, and R^6 are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_x and -L-S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

or R^1 in combination with R^2 forms a fused aromatic or heteroaromatic ring that is optionally sulfonated one or more times, or said ring is substituted by -L-R_X or -L-S_C;

R^3 and R^4 are independently selected from the group consisting of hydrogen, C_1-C_6 alkyl, an aromatic or heteroaromatic ring, L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, hydroxy, or halogen and said aromatic or heteroaromatic ring is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 alkoxy, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

or R^2 in combination with R^3, or R^3 in combination with R^4, forms a 5- or 6-membered alicyclic ring;

R^5 is independently selected from the group consisting of hydrogen, methyl, carboxymethyl, C_2-C_6 alkyl, aryl, heteroaryl, -L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

or R^4 in combination with R^5, or R^5 in combination with R^6, forms a 5- or 6-membered alicyclic ring;

one of E and X is O, S, NR^8, or $CR^{1'}=CR^{2'}$, and the other is absent;

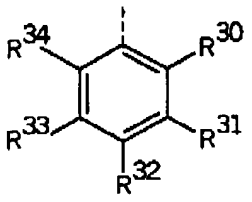
wherein R^8 is independently selected from the group consisting of hydrogen, methyl, carboxymethyl, C_2-C_6 alkyl, -L-R_X and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; and

$R^{1'}$ and $R^{2'}$ are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid, C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

R^{21}, R^{23}, R^{24}, and R^{25} are independently selected from the group consisting of hydrogen, cyano, nitro, halogen, carboxylic acid, sulfonic acid, C_1-C_6 alkyl, aromatic or heteroaromatic ring, -L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, or halogen said aromatic or heteroaromatic ring is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

Q is N or CR^{28}, wherein R^{28} is independently selected from the group consisting of hydrogen, F, CN, carboxylic acid, a carboxylic acid ester of a C_1-C_6 alcohol, a C_1-C_6 alkyl, -L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid,

sulfonic acid, amino, or halogen; or R^{28} comprises a formula



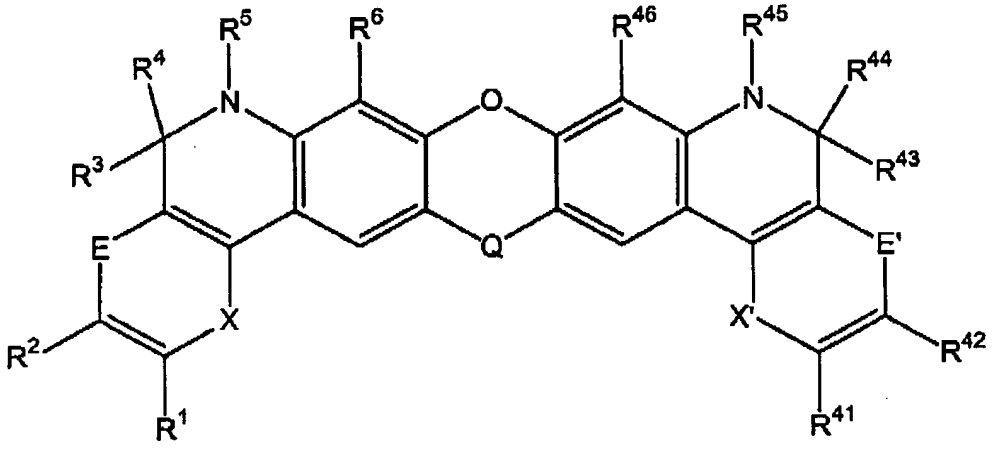
wherein R^{30}, R^{31}, R^{32}, R^{33} and R^{34} are independently selected from the group consisting of hydrogen, F, Cl, Br, I, sulfonic acid, carboxylic acid, CN, nitro, hydroxy, azido, amino, hydrazino, C_1-C_{18} alkyl, C_1-C_{18} alkoxy, C_1-C_{18} alkylthio, C_1-C_{18} alkanoylamino, C_1-C_{18} alkylaminocarbonyl, C_2-C_{36} dialkylaminocarbonyl, C_1-C_{18} alkyloxycarbonyl, C_7-C_{18} arylcarboxamido, -L-R_x and -L-S_C, wherein said alkyl or aryl portions of said R^{30}, R^{31}, R^{32}, R^{33} and R^{34} are optionally substituted one or more times by substituents selected from the group consisting of F, Cl, Br, I, hydroxy, carboxylic acid, a carboxylic acid ester of a C_1-C_6 alcohol, sulfonic acid, amino, C_1-C_6 alkylamino, C_2-C_6 dialkylamino and C_1-C_6 alkoxy; or a pair of adjacent R^{30}, R^{31}, R^{32}, R^{33} and R^{34} substituents when taken in combination, form a fused 6-membered aromatic ring that is optionally further substituted by carboxylic acid; and

wherein L is a covalent linkage;

R_x is a reactive group; and

S_C is a conjugated substance.

22. (Amended) A compound comprising a formula:



wherein R^1, R^2, R^6 R^{41}, R^{42}, and R^{46} are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_x and -L-S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

or R^1 in combination with R^2, or R^{41} in combination with R^{42}, or both, forms a fused aromatic or heteroaromatic ring that is optionally sulfonated one or more times, or said ring is substituted by -L-R_X or -L-S_C;

R^3, R^4, R^{43}, and R^{44} are independently selected from the group consisting of hydrogen, C_1-C_6 alkyl, an aromatic or heteroaromatic ring, L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, hydroxy, or halogen and said aromatic or heteroaromatic ring is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 alkoxy, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

or R^2 in combination with R^3, or R^{42} in combination with R^{43}, or R^3 in combination with R^4, or R^{43} in combination with R^{44}, or any combination thereof, forms a 5- or 6-membered alicyclic ring;

R^5 and R^{45} are independently selected from the group consisting of hydrogen, methyl, carboxymethyl, C_2-C_6 alkyl, aryl, heteroaryl, -L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

R^2 cont.

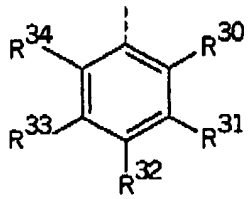
or R^4 in combination with R^5, or R^5 in combination with R^6, or R^{44} in combination with R^{45}, or R^{45} in combination with R^{46}, or any combination thereof, forms a 5- or 6-membered alicyclic ring;

wherein one of said E, E', X' and X is O, S, NR^8, or $CR^{1'}=CR^{2'}$ provided that E and X or E' and X' are not both present;

wherein R^8 is independently selected from the group consisting of hydrogen, methyl, carboxymethyl, C_2-C_6 alkyl, -L-R_X and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; and

$R^{1'}$ and $R^{2'}$ are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid, C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

Q is N or CR^{28}, wherein R^{28} is independently selected from the group consisting of hydrogen, F, CN, carboxylic acid, a carboxylic acid ester of a C_1-C_6 alcohol, a C_1-C_6 alkyl, -L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; or R^{28} comprises a formula



wherein R^{30}, R^{31}, R^{32}, R^{33} and R^{34} are independently selected from the group consisting of hydrogen, F, Cl, Br, I, sulfonic acid, carboxylic acid, CN, nitro, hydroxy, azido, amino, hydrazino, C_1-C_{18} alkyl, C_1-C_{18} alkoxy, C_1-C_{18} alkylthio, C_1-C_{18} alkanoylamino, C_1-C_{18} alkylaminocarbonyl, C_2-C_{36} dialkylaminocarbonyl, C_1-C_{18} alkyloxycarbonyl, C_7-C_{18} arylcarboxamido, -L-R_x and -L-S_C, wherein said alkyl or aryl portions of said R^{30}, R^{31}, R^{32}, R^{33} and R^{34} are optionally substituted one or more times by substituents selected from the group consisting of F, Cl, Br, I, hydroxy, carboxylic acid, a carboxylic acid ester of a C_1-C_6 alcohol, sulfonic acid, amino, C_1-C_6 alkylamino, C_2-C_6 dialkylamino and C_1-C_6 alkoxy; or a pair of adjacent R^{30}, R^{31}, R^{32}, R^{33} and R^{34} substituents when taken in combination, form a fused 6-membered aromatic ring that is optionally further substituted by carboxylic acid; and

B^2 Cont.

wherein L is a covalent linkage;

R_x is a reactive group; and

S_C is a conjugated substance.

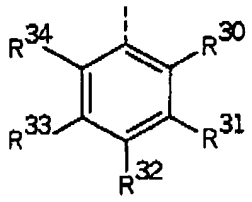
---

23. (Cancel) A compound, as claimed in Claim 22, wherein

X = X', E = E', $R^1 = R^{41}$, and $R^2 = R^{42}$.

R^3

24. (Amended) The compound according to Claim 22, wherein Q is CR^{28} and R^{28} has the formula



B3

25. (Amended) The compound according to Claim 24, wherein one of R^5, R^{30}, R^{31}, R^{32}, R^{33}, R^{34}, and R^{45} is -L-R_x or -L-S_c.

26. (Amended) The compound according to Claim 24, wherein

said R^3, R^4, R^{43}, and R^{44} are each methyl;

each R^1 and R^{41} is independently H or sulfonic acid; and

R^6 and R^{46} are H.

27. (Amended) The compound according to Claim 24, wherein said compound is substituted one or more times by sulfonic acid.

28. (Amended) The compound according to Claim 22, wherein one of said R^1, $R^{1'}$, R^2, $R^{2'}$, R^3, R^4, R^5, R^6, R^{28}, R^{30}, R^{31}, R^{32}, R^{33}, R^{34}, R^{41}, R^{42}, R^{43}, R^{44}, R^{45}, and R^{46} is -L-R_x or -L-S_c.

29. (Amended) The compound according to Claim 28, wherein each L is independently a single covalent bond, or L is a covalent linkage having 1-20 nonhydrogen atoms selected from the group consisting of C, N, O, P, and S.

30. (Amended) The compound according to Claim 28, wherein said R_x is independently selected from the group consisting of an acrylamide, an activated ester of a carboxylic acid, an acyl azide, an acyl nitrile, an aldehyde, an alkyl halide, an amine, an anhydride, an aniline, an aryl halide, an azide, an aziridine, a boronate, a carboxylic acid, a

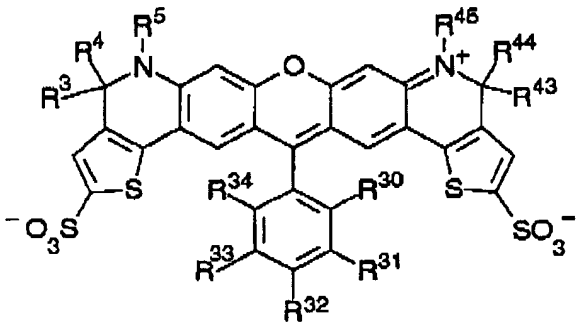
diazoalkane, a haloacetamide, a halotriazine, a hydrazine, an imido ester, an isocyanate, an isothiocyanate, a maleimide, a phosphoramidite, a reactive platinum complex, a sulfonyl halide, a perfluorobenzamido, an azidoperfluorobenzamido group, and a thiol group.

31. (Amended) The compound according to Claim 30, wherein said R_X is independently selected from the group consisting of a phosphoramidite, a succinimidyl ester of a carboxylic acid, a haloacetamide, a hydrazine, an isothiocyanate, a maleimide group, a perfluorobenzamido, an azidoperfluorobenzamido group, and a reactive platinum complex.

32. (Amended) The compound according to Claim 28, wherein said S_C is independently selected from the group consisting of an amino acid, a peptide, a protein, a tyramine, a carbohydrate, an ion-complexing moiety, a nucleoside, a nucleotide, an oligonucleotide, a nucleic acid, a hapten, a psoralen, a drug, a hormone, a lipid, a lipid assembly, a polymer, a polymeric microparticle, a biological cell, and a virus.

33. (Amended) The compound according to Claim 32, wherein S_C is an amino acid, a peptide, a protein, an ion-complexing moiety, a nucleoside, a nucleotide, an oligonucleotide, and a nucleic acid.

34. (Amended) The compound according to Claim 28, wherein said compound comprises a formula:



wherein said R^3, R^4, R^5, R^{43}, R^{44} , and R^{45} are independently methyl or ethyl;
R^{30} is sulfonic acid or carboxylic acid;
R^{31} and R^{34} are independently H, F, or Cl;
one of R^{32} and R^{33} is H, F, or Cl, and the other of R^{32} and R^{33} is -L-R_x or -L-S_c,

wherein said L is a covalent linkage comprising $-S(CH_2)_aCOO(CH_2)_b-$ or $-S(CH_2)_aCONH(CH_2)_b-$

wherein a is an integer between 0 and 10, and b is an integer between 0 and 10; and

wherein said R_x is selected from the group consisting of a carboxylic acid, an activated ester of a carboxylic acid, a haloacetamide, a hydrazine, an isothiocyanate, a maleimide group, and a reactive platinum complex.; and

wherein said S_c is selected from the group consisting of an amino acid, a peptide, a protein, an ion-complexing moiety, a nucleoside, a nucleotide, an oligonucleotide, a lectin, or a nucleic acid.

R3 con.

35. (Amended) The compound according to Claim 34, wherein said R_x is a maleimide group or is a succcinimidyl ester of a carboxylic acid.

36. (Amended) The compound according to Claim 34, wherein said S_c is a peptide or a protein.

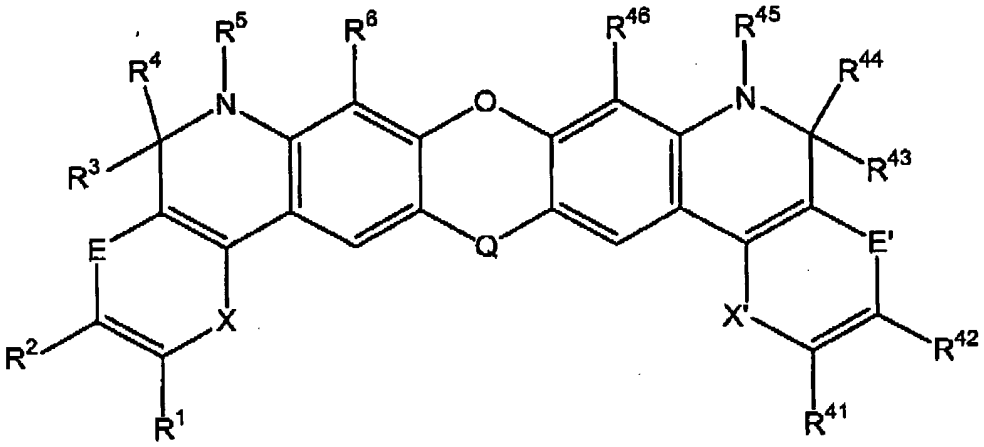
37. (Amended) The compound according to Claim 36, wherein said S_c is an antibody or antibody fragment or a lectin.

38. (Amended) The compound according to Claim 34, wherein said S_c is a nucleotide or an oligonucleotide.

39. (Amended) The compound according to Claim 34, wherein said S_c is a BAPTA or APTRA ion-complexing moiety.

40. (Amended) A method of staining a sample, said method comprising steps:

a) combining a solution with said sample, wherein said solution comprises a compound having formula



wherein R^1, R^2, R^6, R^{41}, R^{42}, and R^{46} are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_x and -L-S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

or R^1 in combination with R^2, or R^{41} in combination with R^{42}, or both, forms a fused aromatic or heteroaromatic ring that is optionally sulfonated one or more times, or said ring is substituted by -L-R_X or -L-S_C;

R^3, R^4, R^{43}, and R^{44} are independently selected from the group consisting of hydrogen, C_1-C_6 alkyl, an aromatic or heteroaromatic ring, L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, hydroxy, or halogen and

said aromatic or heteroaromatic ring is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 alkoxy, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

or R^2 in combination with R^3, or R^{42} in combination with R^{43}, or R^3 in combination with R^4, or R^{43} in combination with R^{44}, or any combination thereof, forms a 5- or 6-membered alicyclic ring;

R^5 and R^{45} are independently selected from the group consisting of hydrogen, methyl, carboxymethyl, C_2-C_6 alkyl, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

B^3 Com.

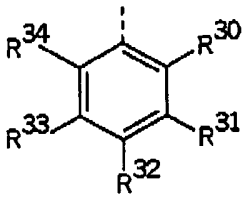
or R^4 in combination with R^5, or R^5 in combination with R^6, or R^{44} in combination with R^{45}, or R^{46} in combination with R^{46}, or any combination thereof, forms a 5- or 6-membered alicyclic ring;

wherein one of said E, E', X' and X is O, S, NR^8, or $CR^{1'}$=$CR^{2'}$ provided that E and X or E' and X' are not both present;

wherein R^8 is independently selected from the group consisting of hydrogen, methyl, carboxymethyl, C_2-C_6 alkyl, -L-R_X and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; and

$R^{1'}$ and $R^{2'}$ are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid, C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

Q is N or CR^{28}, wherein R^{28} is independently selected from the group consisting of hydrogen, F, CN, carboxylic acid, a carboxylic acid ester of a C_1-C_6 alcohol, a C_1-C_6 alkyl, -L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; or R^{28} comprises a formula



wherein R^{30}, R^{31}, R^{32}, R^{33} and R^{34} are independently selected from the group consisting of hydrogen, F, Cl, Br, I, sulfonic acid, carboxylic acid, CN, nitro, hydroxy, azido, amino, hydrazino, C_1-C_{18} alkyl, C_1-C_{18} alkoxy, C_1-C_{18} alkylthio, C_1-C_{18} alkanoylamino, C_1-C_{18} alkylaminocarbonyl, C_2-C_{36} dialkylaminocarbonyl, C_1-C_{18} alkyloxycarbonyl, C_7-C_{18} arylcarboxamido, -L-R_x and -L-S_C, wherein said alkyl or aryl portions of said R^{30}, R^{31}, R^{32}, R^{33} and R^{34} are optionally substituted one or more times by substituents selected from the group consisting of F, Cl, Br, I, hydroxy, carboxylic acid, a carboxylic acid ester of a C_1-C_6 alcohol, sulfonic acid, amino, C_1-C_6 alkylamino, C_2-C_6 dialkylamino and C_1-C_6 alkoxy; or a pair of adjacent R^{30}, R^{31}, R^{32}, R^{33} and R^{34} substituents when taken in combination, form a fused 6-membered aromatic ring that is optionally further substituted by carboxylic acid; and

wherein L is a covalent linkage;

R_x is a reactive group; and

S_C is a conjugated substance;

b) illuminating said sample with a suitable light wavelength to yield a detectable optical response.

B3 CONT.

41. (Amended) The method according to Claim 40, wherein said method further comprises combining said sample with an additional detection reagent.

42. (Cancelled) A method, as claimed in Claim 40, further comprising the step of determining a characteristic of the sample by comparing the optical response with a standard response parameter.

B4

43. (Amended) The method according to Claim 40, wherein said sample comprises cells, growth medium, tissue, proteins, peptides, or biological fluids.

44. (Amended) The method according to Claim 40, wherein said sample is immobilized in or on a solid or semi-solid matrix that is a membrane, an electrophoretic gel, a silicon chip, a glass slide, a microwell plate, or a microfluidic chip.

45. (Cancelled) A method, as claimed in Claim 40, further comprising tracing the temporal or spatial location of the optical response within the sample.

46. (Amended) The method according to Claim 40, wherein at least one of said R^{28}, R^{30}, R^{31}, R^{32}, R^{33}, and R^{34} is -L-R_x or -L-S_C;

B5

R_x is selected from the group consisting of a carboxylic acid, an activated ester of a carboxylic acid, an amine, an azide, a hydrazine, a haloacetamide, an alkyl halide, an isothiocyanate, and a maleimide group; and

S_c is selected from the group consisting of an amino acid, a peptide, a protein, a polysaccharide, a nucleotide, a nucleoside, an oligonucleotide, a nucleic acid polymer, an ion-complexing moiety, a lipid, or a non-biological organic polymer or polymeric microparticle, wherein said Sc is optionally bound to one or more additional fluorophores.

47. (Amended) The method according to Claim 46, wherein said R^{28} is an -L-S_C, and S_C

is an ion-complexing moiety that is a BAPTA or an APTRA.

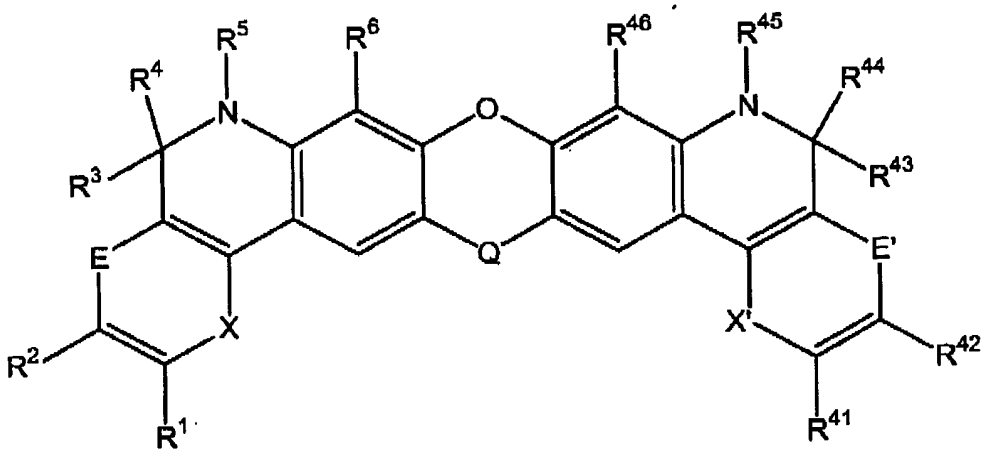
B5 Cont-

48. (Amended) The method according to Claim 46, wherein at least one of said R^{28}, R^{30}, R^{31}, R^{32}, R^{33}, and R^{34} is -L-S_C, and said S_C is a nucleoside, a nucleotide, an oligonucleotide, or a nucleic acid polymer.

49. (New) A kit for staining a sample, wherein said kit comprises

a solution comprising a buffer and a compound having formula

R6



wherein R^1, R^2, R^6, R^{41}, R^{42} and R^{46} are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_x and -L-S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

or R^1 in combination with R^2, or R^{41} in combination with R^{42}, or both, forms a fused aromatic or heteroaromatic ring that is optionally sulfonated one or more times, or said ring is substituted by -L-R_X or -L-S_C;

R^3, R^4, R^{43}, and R^{44} are independently selected from the group consisting of hydrogen, C_1-C_6 alkyl, an aromatic ring, a heteroaromatic ring, L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, hydroxy, or halogen and said aromatic or heteroaromatic ring is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 alkoxy, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

or R^2 in combination with R^3, or R^{42} in combination with R^{43}, or R^3 in combination with R^4, or R^{43} in combination with R^{44}, or any combination thereof, forms a 5- or 6-membered alicyclic ring;

R^5 and R^{45} are independently selected from the group consisting of hydrogen, methyl, carboxymethyl, C_2-C_6 alkyl, aryl, heteroaryl, -L-R_x and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, carboxylic acid, sulfonic acid, or halomethyl;

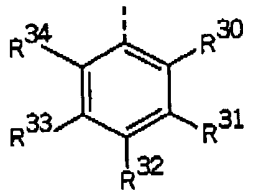
or R^4 in combination with R^5, or R^5 in combination with R^6, or R^{44} in combination with R^{45}, or R^{45} in combination with R^{46}, or any combination thereof, forms a 5- or 6-membered alicyclic ring;

wherein one of said E, E', X' and X is O, S, NR^8, or $CR^{1'}=CR^{2'}$, provided that E and X or E' and X' are not both present;

wherein R^8 is independently selected from the group consisting of hydrogen, methyl, carboxymethyl, C_2-C_6 alkyl, -L-R_X and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; and

$R^{1'}$ and $R^{2'}$ are independently selected from the group consisting of hydrogen, cyano, halogen, carboxylic acid, sulfonic acid, C_1-C_6 alkyl, C_1-C_6 alkoxy, aryl, heteroaryl, -L-R_X and -L-S_C, wherein said alkyl or alkoxy is optionally substituted by carboxylic acid, sulfonic acid, or halogen and said aryl or heteroaryl is optionally substituted one or more times by C_1-C_6 alkyl, C_1-C_6 perfluoroalkyl, cyano, halogen, azido, carboxylic acid, sulfonic acid, or halomethyl;

Q is N or CR^{28}, wherein R^{28} is independently selected from the group consisting of hydrogen, F, CN, carboxylic acid, a carboxylic acid ester of a C_1-C_6 alcohol, a C_1-C_6 alkyl, -L-R_X and -L-S_C, wherein said alkyl is optionally substituted by carboxylic acid, sulfonic acid, amino, or halogen; or R^{28} comprises a formula



wherein R^{30}, R^{31}, R^{32}, R^{33} and R^{34} are independently selected from the group consisting of hydrogen, F, Cl, Br, I, sulfonic acid, carboxylic acid, CN, nitro, hydroxy, azido, amino, hydrazine, C_1-C_{18} alkyl, C_1-C_{18} alkoxy, C_1-C_{18} alkylthio, C_1-C_{18} alkanoylamino, C_1-C_{18} alkylaminocarbonyl, C_2-C_{36} dialkylaminocarbonyl, C_1-C_{18} alkyloxycarbonyl, C_7-C_{18} arylcarboxamido, -L-R_X and -L-S_C, wherein said alkyl or aryl portions of said R^{30}, R^{31}, R^{32}, R^{33} and R^{34} are optionally substituted one or more times by substituents selected from the group consisting of F, Cl, Br, I, hydroxy, carboxylic acid, a carboxylic acid ester of a C_1-C_6 alcohol, sulfonic acid, amino, C_1-C_6 alkylamino, C_2-C_6 dialkylamino and C_1-C_6 alkoxy; or a pair of adjacent R^{30}, R^{31}, R^{32}, R^{33} and R^{34} substituents when taken in combination, form a fused 6-membered aromatic ring that is optionally further substituted by carboxylic acid; and

wherein L is a covalent linkage;

R_x is a reactive group; and

S_c is a conjugated substance.

50. (New) The kit according to Claim 49, wherein said kit further comprises an additional detection reagent, a purification medium, or standards.

51. (New) The kit according to Claim 49, wherein at least one of said R^1, R^2, R^3, R^4, R^5, R^6, R^{28}, R^{30}, R^{31}, R^{32}, R^{33}, R^{34}, R^{43}, R^{44}, R^{45} and R^{46} is L-R_x wherein said R_x is independently selected from the group consisting of a carboxylic acid, an activated ester of a carboxylic acid, an amine, an azide, a hydrazine, a haloacetamide, an alkyl halide, an isothiocyanate, and a maleimide group.

52. (New) The kit according to Claim 51, wherein at least one of said R^{31}, R^{32}, R^{33}, or R^{34} is L-R_x and R^{30} is carboxylic acid or sulfonic acid.

53. (New) The kit according to Claim 49, wherein at least one of said R^1, R^2, R^3, R^4, R^5, R^6, R^{28}, R^{30}, R^{31}, R^{32}, R^{33}, R^{34}, R^{43}, R^{44}, R^{45} and R^{46} is L-Sc, wherein said Sc is independently selected from the group consisting of an amino acid, a peptide, a protein, an antibody, an antibody fragment, a carbohydrate, a nucleotide, a nucleoside, an oligonucleotide, a nucleic acid polymer, an ion-complexing moiety, a lipid, a non-biological organic polymer and polymeric microparticle.

54. (New) The kit according to Claim 53, wherein said Sc is an antibody or fragment thereof.

Diwu *et al.*
Serial No. 09/922,333

Docket No. D305.001PN.2

Respectfully submitted,

Date: December 19, 2002

Koren J. Anderson

Koren J. Anderson, PhD
Reg. No. 51,081

Molecular Probes, Inc.
P.O. Box 22010
Eugene, Oregon, 97402
Phone: (541) 984-5656
Facsimile: (541) 465- 8354

AUTOMATIC COVER SHEET

DATE: DEC-19-2002 THU 12:02 PM

TO:

FAX #: 917038729306

FROM: MOLECULAR PROBES INC

FAX #: 15414658354

70 PAGES WERE SENT

(INCLUDING THIS COVER PAGE)

Received from < 15414658354 > at 12/19/02 2:40:01 PM [Eastern Standard Time]